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TITLE: Granulopoietic Growth Factor Secretion in Ovarian Carcinoma as a Mechanism for the Emergence of Immune Suppressive Myeloid Subsets

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Fort Detrick, Maryland 21702-5012

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14. ABSTRACT A major problem facing effective ovarian cancer immunotherapy is the capacity of malignant populations to inhibit immune activation or evade destruction. Among prominent mechanisms thought to impede the anticancer response is the accumulation of pro-tumorigenic myeloid populations, termed myeloid-derived suppressor cells (MDSC). However, there is a fundamental gap in knowledge regarding the exact mechanisms that drive their development or accumulation. To that end, we originally hypothesized that the overproduction of granulocyte-colony stimulating factor (G-CSF) in ovarian carcinoma facilitates MDSC accumulation. However, during the course of these studies, we discovered that the levels of IL-8 overwhelmed all other cytokines/chemokines tested, including G-CSF. Consequently, we refined our original hypothesis to reflect tumor-derived IL-8, in lieu of G-CSF as a major component of MDSC-mediated ovarian tumor progression. Thus far, our data have led to the: *1)* identification of distinct MDSC subsets in ovarian cancer patients; *2)* identification of several pro-inflammatory cytokines in matched patient sera, notably IL-8; *3)* development of a xenograft model that recapitulates the MDSC and IL-8 phenotype seen in patient samples; and *4)* observation that IL-8 knockdown in ovarian cancer cell line models significantly improves overall survival. Altogether, this new axis in myeloid-ovarian tumor biology has important implications for IL-8-based clinical interventions.

15. SUBJECT TERMS
ovarian cancer, myelopoietic growth factors, myeloid suppressor cells, immune suppression, tumor progression, immunotherapy

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Table of Contents

1. Introduction: We now know that myeloid-derived suppressor cells (MDSC) comprise immature myeloid populations produced in a wide range of human cancers, including ovarian carcinoma, and play significant tumor-promoting roles by suppressing adaptive immunity and by providing a rich source of angiogenic factors [1]. We also know that MDSC are composed of two major subsets, monocytic and granulocytic, the proportion of each is influenced by the types of tumor-derived factors (TDF) secreted *in vivo* [1,2]. Although much attention has focused on understanding how MDSC function, a larger gap remains in our understanding of mechanisms that govern their development or recruitment to the tumor microenvironment. To that end, our original proposal addressed the novel concept that overproduction of granulocyte-colony stimulating factor (G-CSF) in ovarian carcinoma facilitates granulocytic MDSC accumulation. However, during the course of these studies, we serendipitously discovered that the levels of IL-8 (CXCL8) overshadowed all other cytokines/chemokines tested, including G-CSF. IL-8, unlike G-CSF, also possesses potent chemoattractant and angiogenic properties [3]. Given this striking new finding, *we refined our original hypothesis to reflect tumor-derived IL-8, in lieu of G-CSF as a major element of MDSC-mediated ovarian tumor progression*. The following is a detailed account of our progress made for each aim and task outlined in the original SOW.

2. Body (from Original SOW):

Specific Aim 1*: To quantify G-CSF levels in human ovarian cancer cell lines, and then evaluate the ability of selected G-CSF-producing cell lines to generate MDSC using human-mouse xenograft models*

***Task 1a*:** Cell-free supernatants of human ovarian cancer cell lines will be quantified for G-CSF levels. Both metastatic SKOV-3 and N IH:OVCAR-3 cell lines (from ATCC) will be analyzed, since both have been shown to express G-CSF *(timeframe, months 1-2)*

Progress: Completed, and determined that both cell lines produce G-CSF as expected. However, the concentrations were also lower than expected. Therefore, before pursuing subsequent studies, we embarked on a more comprehensive analysis of tumor-derived cytokines and chemokines, focusing on two key characteristics: association with MDSC biology and cross-reaction in human-mouse xenograft models. In doing so, we compiled a list of 12 distinct cytokines and chemokines, including G-CSF and analyzed them by Luminex Multiplex technology (Fig. 1A and 1B). First, our findings confirmed G-CSF production at similar levels determined by ELISA (< 100 pg/ml). Secondly, we identified an additional five factors that were highly produced, most notably IL-8 which was common to both cell lines. In fact, IL-8 levels exceeded 2 ng/ml/10^6 cells/24 hr.

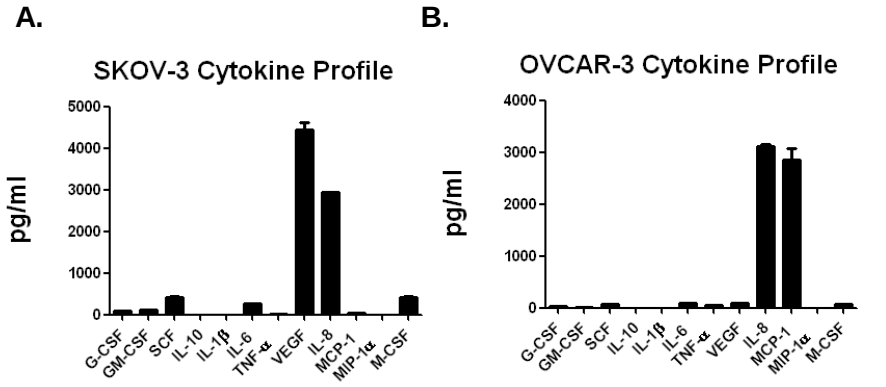

Figure 1. Cytokine production by ovarian carcinoma cell line models. Cell-free culture supernatants were analyzed by Luminex Multiplex technology for the indicated TDF. D ata reported as pg/ml/10^6 cells/24 hr.

Given the striking finding with IL-8, we extended this analysis to patient sera (Fig. 2), which is described further in Aim 3. Briefly, similar to what we observed with both cell lines, we found copious levels of IL-8 in a majority of patient sera. Also, as with both cell lines, we identified five additional factors, several of which were shared with the tumor cell lines. Importantly, all six factors were significantly higher compared to those of age- and race-matched healthy female donors, suggesting that TDF differences between case and control were due to the neoplastic process. Thus, the high degree of similarity between cell line and patient data provided validation for continued use of the cell line model. Altogether, while these data supported our original concept, we opted to pursue IL-8 over G-CSF. We remain cognizant of the fact that five remaining TDF are available to study in future studies, alone or in combination with IL-8.

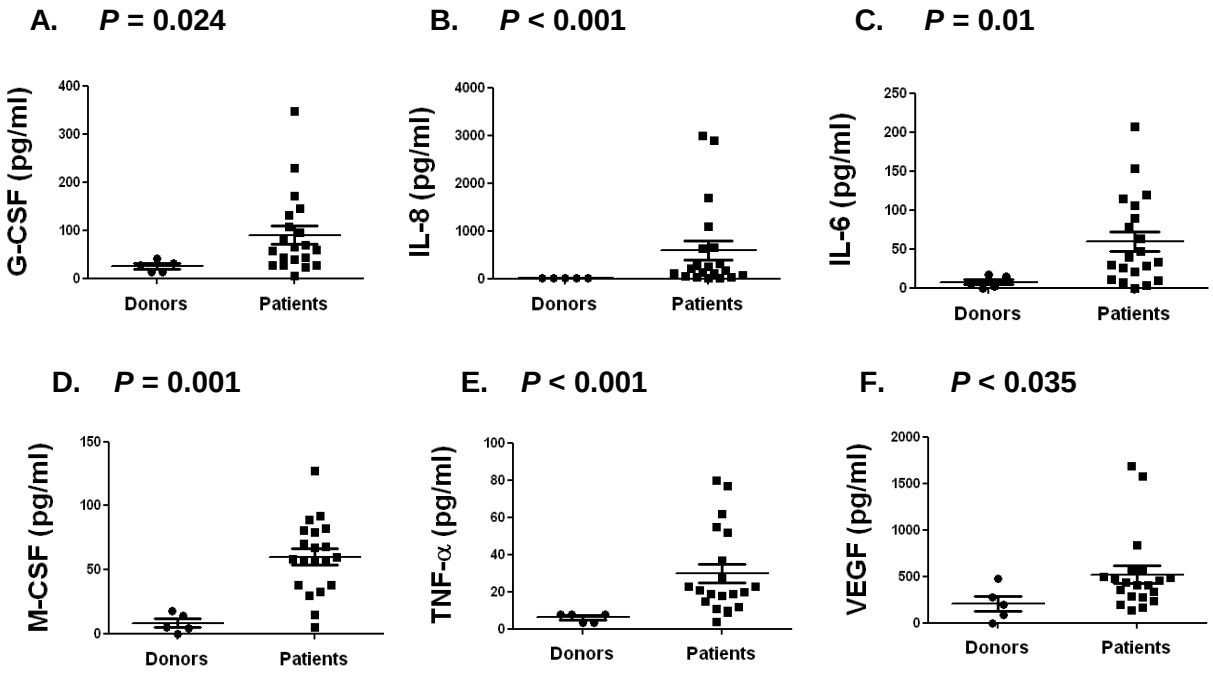

Figure 2. Serum cytokine levels in healthy vs. ovarian cancer patients. Serum from the indicated cohorts analyzed as in Fig. 1. Each point reflects an individual donor or patient. All patient data shown are significant from age- and race-matched donors. Six additional factors tested showed no significant differences in relation to controls. GM-CSF, SCF, IL-1β, IL-10,MCP-1, MIP-1α.

Task 1b: RNA will be collected from the cells to verify G-CSF message levels by RT-PCR or real-time PCR.

Progress: Since our protein data was most revealing, we did not pursue molecular analysis of G-CSF in our cell line model, but confirmed IL-8 message, as described further in Aim 2.

Task 2a: Evaluate the ability of ovarian tumor cell lines to produce G-CSF and generate granulocytic MDSC *in vivo* (*timeframe, months, 3-6*)

Progress: We followed through with orthotropic implantation of each cell line. Importantly, both cell lines were tumorigenic *in vivo*; however, SKOV3 seemed to be more tumorigenic than NIH:OVCAR-3, based on time to morbidity (Fig. 3). Therefore, since SKOV3 was more aggressive, we opted to pursue this cell line first in subsequent experiments.

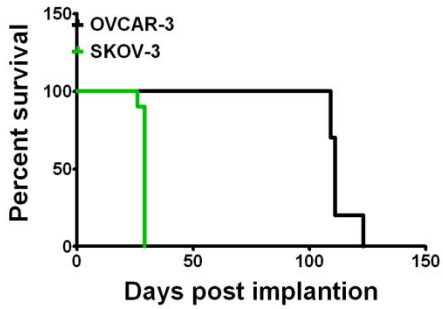

Figure 3. Human-mouse xenograft model of ovarian tumor growth. Kaplan-Meier plots of the indicated tumor-bearing mice. Tumor cells were implanted *ip* ($5x10^6$/mouse) and mice euthanized immediately upon signs of morbidity (n = 10/group).

Task 2b: Both sera G-CSF and MDSC levels will be determined and tracked at biweekly intervals during the course of tumor growth, based on blood draw. Age/gender-matched non-tumor-bearing mice will serve as controls for G-CSF and MDSC values at all measurable time-points.

Progress: Based on our results in Task 1, we plan to analyze sera for IL-8 levels. Currently, sera is banked, but we did collect data for IL-8 levels in Aim 2 (see below). However, in regard to MDSC quantification, we focused on the spleen as a systemic reservoir for the accumulation of MDSC during tumor-bearing conditions. Data is discussed below.

Task 2c: When mice appear moribund (as evidence of advanced disease), mice will be euthanized and spleens and ascites (tumor site) collected. Blood will also be collected as part of a terminal bleed. Single cell suspensions will be made from splenic and tumor sites, and cryopreserved for subsequent studies.

Progress: Completed; all cells and tissues collected as proposed.

Task 2d: Cell preparations above will be thawed. Murine granulocytic MDSC will be defined by their $CD11b^+Ly6C^{lo}Ly6G^+$ phenotype by flow cytometry, granulocytic morphology by Wright-Giemsa staining and ability to inhibit T cell proliferation using *in vitro* assays described in Fig. 2.

Progress: Analysis of MDSC frequencies in the spleen revealed significant levels of MDSC, particularly of the granulocytic phenotype, relative to the non-tumor-bearing control mice (Fig. 4). This was observed in both tumor cell line models, indicating that ovarian carcinoma can generate MDSC *in vivo*. Morphologic studies in an independent model of mammary carcinoma confirmed that the MDSC expressing this phenotype is granulocytic. T cell proliferation studies are pending.

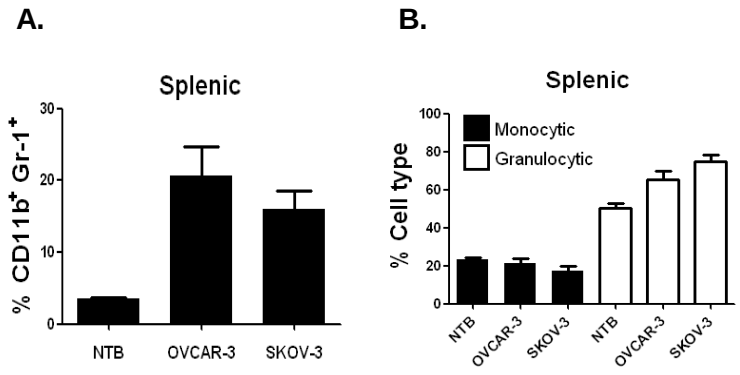

Figure 4. MDSC formation during ovarian tumor growth. At endpoint (Fig. 3), spleens were collected and analyzed individually for MDSC, based on the global population (*A*), as well as specific myeloid subsets (*B*). Monocytic, $CD11b^+Ly6G^-Ly6C^{hi}$; granulocytic, $CD11b^+Ly6G^+Ly6C^{lo}$ (n = 3 of 10 analyzed).

Milestones for Aim 1: These studies will identify relevant ovarian tumor cell line models that satisfy the scientific criteria for G-CSF production and granulocytic MDSC generation *in vivo*.

Overall Progress for Aim 1: We have completed major elements of this aim. First, we identified ovarian tumor cell lines that are tumorigenic in this *in vivo* model. Secondly, tumor growth was accompanied by a significant MDSC response, largely due to the accumulation of the granulocytic subset as originally hypothesized. And thirdly, while we did confirm tumor-derived G-CSF production, we observed that the G-CSF response paled in comparison to IL-8 production in both cell line models and patient sera. This serendipitous observation thus provided the rationale to explore IL-8 instead of G-CSF in Aim 2. Based on our preliminary data in Aim 2, we believe this observation has a strong likelihood of success. Also, it is very important to note all further studies below remain essentially unchanged, and will be conducted with an IL-8 instead of G-CSF focus.

Specific Aim 2: *To examine the causal link between tumor-derived G-CSF production and granulocytic MDSC development using loss-of-function approaches*

Task 1a: To generate ovarian tumor cell lines in Aim 1 deficient in G-CSF production by shRNA-based methodologies via our shRNA core facility *(timeframe, during months 7-10):*

Progress: Based on our data in Aim 1, we replaced G-CSF with IL-8. At this time, we decided to focus on one cell line, SKOV3, due to its ability to grow more reliably and aggressively than NIH:OVCAR-3 (Fig. 3). The various sublines have been successfully generated.

Task 1b: Analyze and verify the efficiency of gene knockdown in the different populations via real-time PCR analysis and ELISA.

Progress: Completed; characterization of the different sublines revealed varying degrees of IL-8 knockdown relative to the scramble control (Fig. 5A). The magnitude of knockdown was confirmed by ELISA (Fig. 5B). Based on these criteria, we selected construct 1 for subsequent studies.

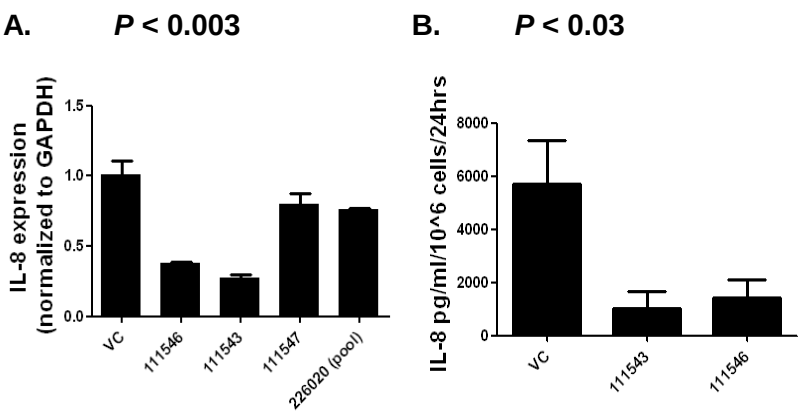

Figure 5. Generation of IL-8-deficient ovarian tumor sublines. SKOV3 cells were silenced for IL-8, based on shRNA methods. VC refers to the scramble control, while each numbered code represents a given subline expressing a single construct or a pool of all 3. Data analyzed by qPCR (*A*) or ELISA (*B*) for secreted protein for the VC and the two sublines showing the highest knockdown in *A*.

Task 1c: For each tumor cell line, identify and select the population most deficient in G-CSF expression. Characterize each further for potential off-target biologic effects, such as changes in cell proliferation *in vitro* by ^{3}H-thymidine assays and expression of MHC class I molecules by flow cytometry.

Progress: IL-8 knockdown did not alter in vitro proliferation (Fig. 6A). Effects on class I expression are pending.

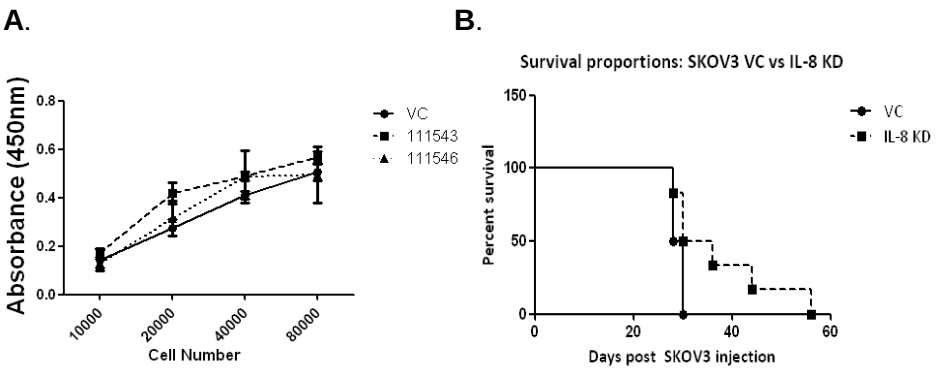

Figure 6. Effect of IL-8 knockdown on tumor cell growth *in vitro* or *in vivo*. (*A*) Cells were plated at varying densities and proliferation measured *in vitro* by a MTT-based assay. (*B*) Orthotopic tumor growth in female SCID mice, as in Fig. 3 (n = 11 for VC and 6 f or 111543).

Milestones for Task 1: Development and characterization of human ovarian tumor cell lines that are stably silenced for G-CSF production. For each tumor cell line, two populations will be generated, one deficient in G-CSF and the other serving as the vector control.

Progress: We have also achieved major elements of this aim. Importantly, we have produced both control and IL-8-deficient sublines for subsequent studies.

Task 2a: To evaluate the effect of G-CSF-deficiency on t umor growth; collection of serum, lymphoid and tumor tissues from mice for subsequent phenotypic and functional analyses *(timeframe, during months 11-13)*:

Progress: Completed for the SKOV3 model.

Task 2b: For proof-of-concept, emphasis will be on the endpoint analysis. When mice appear moribund as evidence of advanced disease, mice will be euthanized and splenocytes, ascites and serum collected and stored as in Aim 1/Task 2.

Progress: Completed for the SKOV3 model. Interestingly, we observed a s ignificant increase in overall survival in mice bearing the IL-8-deficient subline compared to the scramble control (Fig. 6B above).

Milestones for Task 2: Determination of the impact of G-CSF-deficiency on orthotopic tumor growth using two different ovarian tumor models.

Progress: Completed for the SKOV3 model. Altogether, these data are promising and suggest an important role of tumor-derived IL-8 production in ovarian cancer progression.

Task 3a: To analyze serum G-CSF levels and MDSC accumulation in both lymphoid and tumor tissues (Experiments will be staggered so the different tumor models will be analyzed at different times) (*timeframe, during months 14-16*):

Progress: Completed for the SKOV3 model. First, we observed a significant difference in IL-8 levels, pre- vs. post-tumor challenge in mice bearing either the scramble control or IL-8-deficient subline. Whereas, pre-bleeds showed no detectable IL-8, post-bleeds revealed significant amounts of circulating IL-8 (Fig. 7A and 7B). Importantly, the level of IL-8 in sera of

mice bearing the IL-8-deficient subline was significantly lower compared to the scramble control (Fig. 7C). The post-bleeds were taken at endpoint when all mice appeared moribund. These data not only validate the IL-8 phenotype in vivo, but also demonstrate a correlation with survival.

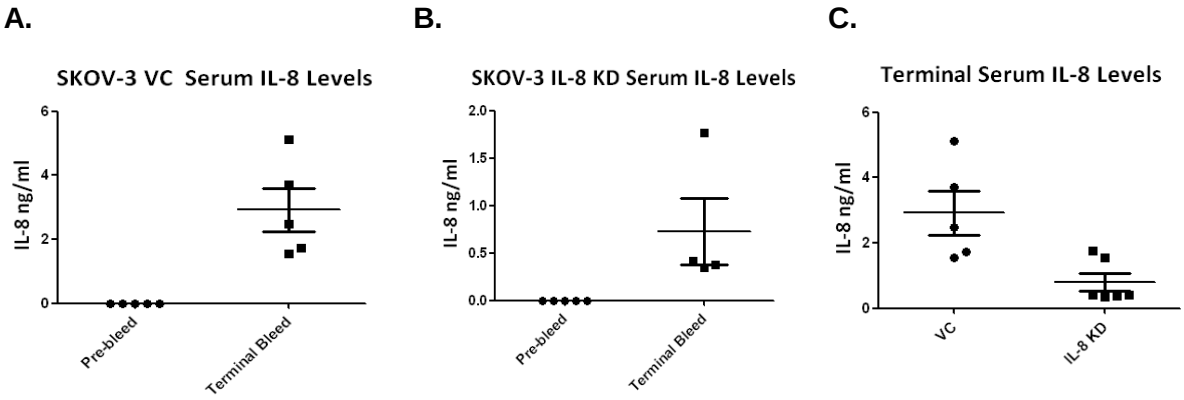

Figure 7. Serum IL-8 levels during orthotopic tumor growth. (*A, B*) IL-8 levels pre-implantation and post-tumor implantation at endpoint for the indicated sublines. Serum isolated from mice shown in Fig. 6 (IL-8 KD; construct 111543). (*C*) IL-8 levels at endpoint, comparing VC to IL-8-deficient cells ($P < 0.05$). Each data point in *A-C* denotes a single mouse measurement.

Task 3b: Thaw splenocytes, and anal yze for granulocytic MDSC levels (percentages/absolute numbers) based on $CD11b^{+}Ly6C^{lo}Ly6G^{+}$ phenotype by flow cytometry.

Task 3c: Purify splenic granulocytic MDSC using magnetic bead cell separation kits and identify/verify cellular morphology based on Wright-Giemsa staining.

Task 3d: Assess purified myeloid cells for their ability to activate/inhibit T cell activity *in vitro* (via ^{3}H-thymidine assays).

Task 3e: Analyze single cell suspensions from ascites for MDSC characteristics, as in Aim 3/Tasks 3b -3d.

Progress: Experiments in Tasks 3b - 3e are pending.

Task 4a: To analyze the effect of G-CSF blockade on M DSC accumulation using a neutralizing anti-G-CSF monoclonal antibody (mAb) (*timeframe, during month 17-19*):

Task 4b: Spleens and t umor tissues will be c ollected 24 – 48 hr after the final mAb injection. MDSC levels/function will then be assessed as in Aim 2/Task 3.

Progress: Studies are pending for tasks 4a and 4b.

Milestones for Tasks 3, 4 & Overall Aim 2: Determination of the impact of G-CSF-deficiency on tumor growth and MDSC development using two ovarian tumor models. These studies have the potential to identify for the first time that tumor-derived G-CSF exhibits pro-tumorigenic activity, namely the induction of granulocytic MDSC, and t hat strategies that target G-CSF may have novel predictive or therapeutic value.

Overall Progress for Aim 2: Thus far, our data support the hypothesis that tumor-derived IL-8 plays an important role in ovarian carcinoma progression, and establish the basis for continuation of these studies during year 2.

Specific Aim 3: *To measure circulating G-CSF and MDSC levels in ovarian carcinoma patients and then correlate potential changes in both parameters with clinical outcome measurements*. Studies will overlap with prior Aims *(timeframe, during months 3-24):*

Task 1: To quantify serum G-CSF levels in 30 ovarian cancer patients with advanced disease (e.g., stages III/IV); compare with 30 age/gender/race-matched healthy female donors.

Progress: Completed for cancer patients (n=21) and m atched controls (n=5). Comprehensive cytokine/chemokine analysis revealed production not only of G-CSF, but robust levels of IL-8, compared to healthy controls (Fig. 2 above). Since the data for the five controls were very tight, we reasoned that it was not necessary to test the remaining samples at this time. Altogether, patient data were consistent with our cell line data.

Task 2: To measure frequencies of granulocytic MDSC in the peripheral blood of the same patients in Aim 3/Task 1:

Progress: Completed. Using multi-parameter flow cytometry, we compared 21 ovarian cancer patients reflecting stages II - IV to 22 gender- race- and age-matched controls. All patient peripheral blood samples were obtained at diagnosis through the Data Bank and Biorepository, and the mononuclear fraction was cryopreserved. We reasoned that patients with advance disease, such as stages II – IV, would best illustrate the relationship between MDSC frequencies in health vs. disease. Our data clearly demonstrated significant levels of all three MDSC subsets in patients compared to phenotypically matched cells from healthy donors. Altogether, these data support the relationship between MDSC levels and disease status. Clinical correlations are pending.

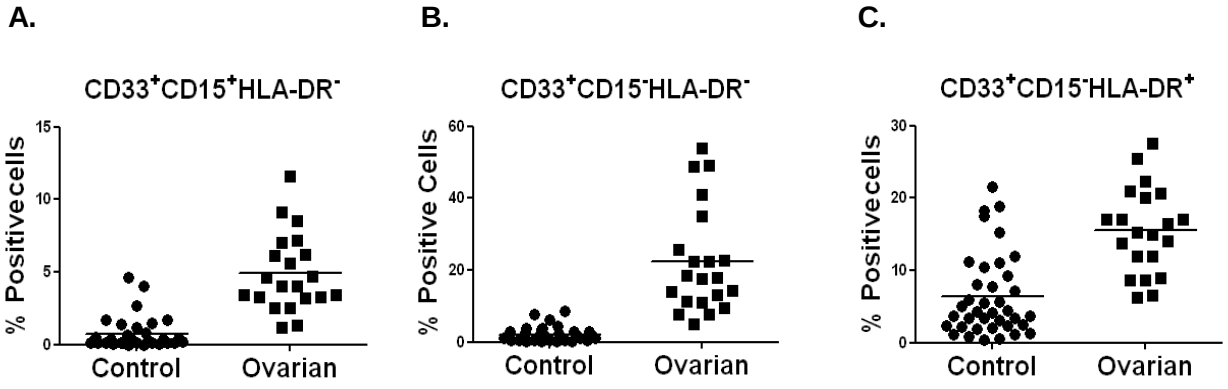

Figure 8. MDSC subset analysis in ovarian cancer patients. Using multi-parameter flow cytometry, 21 ovarian cancer patients reflecting stages II - IV to 22 were compared to race- and age-matched female controls. All patient peripheral blood samples were obtained at diagnosis through the Data Bank and Biorepository at RPCI through a de -identified IRB protocols, and the mononuclear fraction was cryopreserved. Data demonstrate significant levels of all three MDSC subsets in patients compared to phenotypically matched cells from healthy donors ($P < 0.01$ in all three panels). Each data point reflects a single patient or donor.

Task 3: If the data are statistically meaningful in Aim 3/Tasks 1 & 2, patients with earlier stage disease (n=30) will be analyzed in a similar fashion.

Progress: Studies are pending.

Milestones for Aim 3: Sera G-CSF and circulating granulocytic MDSC frequencies will be correlated to each other and against known clinical outcome measurements (CA-125 levels, stage, grade, invasiveness, metastatic location/burden, progression-free or overall survival). *The overall predictions are that G-CSF levels will: 1) correlate with granulocytic MDSC frequencies*; and *2) associate with poorer clinical outcome measurements*. *Thus, these studies may provide the rationale to target G-CSF for prognostic or therapeutic purposes in at least subsets of patients*.

Overall Progress for Aim 3: Thus far, we have identified 6 cytokines/chemokines highly expressed in ovarian cancer patients compared to healthy donors. We are focusing on IL-8, as this cytokine/chemokine overshadowed all others tested in patients, as well as two ovarian tumor cell line models in which to test causation. Concurrent with these studies, we demonstrated that ovarian cancer patients harbor significant frequencies of three distinct MDSC subsets in circulation compared to matched donors. Given that we have identified significant hematologic differences in healthy vs. disease, we will next determine whether: 1) IL-8 levels correlate with MDSC frequencies; and 2) either one or both biologic endpoints correlate with clinical outcome using well-annotated de-identified patient data.

3. Key Research Accomplishments:

- Identification of 3 distinct MDSC subsets in patients with ovarian carcinoma reflecting stages II – IV.
- Identification of six pro-inflammatory cytokines in matched patient sera, including IL-8, G-CSF, M-CSF, IL-6, TNF-α and VEGF-A (all relative to healthy donors). Correlations between cytokine levels, MDSC frequencies and clinical outcome are pending.
- Development of a human-mouse xenograft model that recapitulates the MDSC and IL-8 phenotype seen in patient samples.
- IL-8 knockdown in an ovarian cancer cell line model significantly improves overall survival, which correlates with a concomitant reduction in serum IL-8 levels

4. Reportable Outcomes: Not applicable during this reporting period.

5. Conclusions: The identification of this new axis in MDSC-ovarian tumor biology has important implications for IL-8-based clinical interventions. Monitoring changes in IL-8 levels, systemically or within the tumor microenvironment, along with other clinical parameters, may serve as a novel 'biomarker' signature for disease status or response to anticancer therapy.

6. References:

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2. Youn, J.I., Nagaraj, S., Collazo, M. & Gabrilovich, D.I. Subsets of myeloid-derived suppressor cells in tumor-bearing mice. *J Immunol* **181**, 5791-5802 (2008).

3. Waugh, D.J. & Wilson, C. The interleukin-8 pathway in cancer. *Clin Cancer Res* **14**, 6735-6741 (2008).

7. Appendices: Updated Curriculum Vitae

8. Supporting Data: Figures are embedded.

CURRICULUM VITAE

Name: Scott I. Abrams, Ph.D.

Citizenship: United States

Education

1981	B.S. (Biology), Delaware Valley College, Doylestown, Pennsylvania *summa cum laude*
1987	Ph.D. (Microbiology & Immunology), Indiana University School of Medicine, Indianapolis, Indiana

Chronology of Employment

1981 – 1987	Graduate Student, Department of Microbiology and Immunology, Indiana University School of Medicine, Indianapolis, IN
1987 – 1991	Postdoctoral Fellow, Department of Molecular Biology and Pharmacology, Washington University School of Medicine, St. Louis, MO
1991 – 8/15/98	Senior Staff Fellow, Laboratory of Tumor Immunology and Biology, Division of Basic Sciences, National Cancer Institute, National Institutes of Health, Bethesda, MD
8/16/98 – 1/31/08	Investigator, Laboratory of Tumor Immunology and Biology, Center for Cancer Research, National Cancer Institute, National Institutes of Health, Bethesda, MD
2/1/08 – present	Associate Member, Department of Immunology & Associate Professor of Oncology, Roswell Park Cancer Institute, Buffalo, NY

Societies

American Association for Cancer Research
The American Association of Immunologists
The American Association for the Advancement of Science
International Society for Biological Therapy of Cancer
Sigma Xi, The Scientific Research Socieity

Honors, Awards and Other Professional Activities

1987	Nominated by Dissertation Committee and Faculty for the Esther L. Kinsley Dissertation Award, Indiana University's Highest Honor

1987 – 1990	Recipient of National Research Service Awards for Postdoctoral Studies
1994 & 1996	Finalist in Research Competition in the Presidential Session of the Society for Biological Therapy Conference on two separate occasions
1996	NIH Federal Technology Transfer Award
1998	NIH Federal Technology Transfer Award
2000	FY 2000 Intramural Research Award, CCR, NCI, NIH
2000	"On-The-Spot Award", LTIB, CCR, NCI
2001	"On-The-Spot Award", LTIB, CCR, NCI
2002	"On-The-Spot Award", LTIB, CCR, NCI
2003	NIH Federal Technology Transfer Award
2003	Performance Award, LTIB, CCR, NCI
2004	Nominated for 2004 NCI Outstanding Mentor Award
2004	NIH Federal Technology Transfer Award
2004	Performance Award, LTIB, CCR, NCI
2005	Performance Award, LTIB, CCR, NCI
2006	NIH Federal Technology Transfer Award
2006	Performance Award, LTIB, CCR, NCI
2007	NIH Federal Technology Transfer Award
2007	Performance Award, LTIB, CCR, NCI
2011	Recipient of 2011 AAI Laboratory Travel Grant
2012	Recipient of 2012 AAI Laboratory Travel Grant

Leadership Positions at Conferences/Editorial Level

2004	Co-Chair Cancer Vaccine/Immunotherapy Block Symposium at the Experimental Biology Conference, Washington, D.C.
2005	Section Editor for "Drug Discovery Today: Disease Mechanisms". Issue dedicated to "Immune Mechanisms of Cancer"
2010	Co-Chair for the 10th Annual Buffalo Conference on Immunology
2011	Co-Chair, Tumor Immunology mini-symposium at AAI conference
2011 – present	Editorial Board member of Immunological Investigations
2012	Co-Chair, Tumor Immunology mini-symposium at AAI conference
2012	Guest Editor for Immunological Investigations for thematic Issue on "Regulator Myeloid Cells in Neoplastic Disease"

Patents

2002	European Patent #97 921 247.9 - Mutated *ras* Peptides for Generation of CD8$^+$ Cytotoxic Lymphocytes
2010	U.S. Patent #7,709,002 – Mutated *ras* Peptides for Generation of CD8+ Cytotoxic Lymphocytes

Committees and Boards

Member of Curriculum Committee, Graduate Program/Department of Immunology, RPCI
Member of Gene Targeting/Transgenic Steering Committee, RPCI
Member, Tumor Immunology and Immunotherapy Program, RPCI
Member, Institutional Divisional Committee, RPCI

Member, Immunology Steering Committee, RPCI
Chair, Progress Committee (Dept. of Immunology), RPCI
Ad hoc member for Scientific Review Committee, RPCI

Study Sections

Ad hoc reviewer for Welcome Trust
Ad hoc reviewer for Institutional Research Grants from ACS at RPCI
Ad hoc reviewer for Association for International Cancer Research
Ad hoc reviewer for NIH study section, NCI-I (career investigator awards)
Ad hoc reviewer for VA Merit Review Panel

Reviewer

Cancer Research
Clinical Cancer Research
Immunological Investigations
Journal of Immunology
Journal of Immunotherapy
Molecular Cancer Therapeutics
Oncogene
PLoS ONE

Mentorship

Primary Mentor for Jeremy Waight at SUNY-Buffalo/RPCI, who was
- Selected for oral presentation at annual AAI conference, 2011
- Awarded first place in RPCI-Graduate Student Poster Presentation Competition, 2011
- Awarded second place for poster presentation at 11th Annual Buffalo Conference on Immunology, 2011

Primary Mentor for Debarati Banik at SUNY-Buffalo/RPCI, who was
- Selected for oral presentation at annual Upstate New York Immunology Conference,
- Selected for oral presentation at annual AAI conference, 2012

Served/serving on 10 Ph.D. and 2 Master Degree Thesis Committees (Two Ph.D. students recently graduated)

Preceptor for a Postdoctoral Fellow (T. Stewart) who received a NIH Fellows Award for Research Excellence (FARE), 2007

Preceptor for a Postdoctoral Fellow (K. Greeneltch) who received a NIH Fellows Award for Research Excellence (FARE), 2007

Preceptor for a Postdoctoral Fellow (T. Stewart), who received an AACR Scholar-in-Training Award at the AACR 97th Annual Meeting, 2006

Preceptor for a Postdoctoral Fellow (T. Stewart), who was selected for oral presentation at the NCI-CCR Fellow's Retreat, 2006

Preceptor for a Research Fellow (K. Liu), who was selected for oral presentation at the NIH Immunology Retreat, 2005

Preceptor for a Postdoctoral Fellow (T. Stewart), who was selected for oral presentation at the Combined Faculty Retreat, CCR-NCI, 2005

Preceptor for a Research Fellow (K. Liu), whose studies resulted in recognition with a competitive Travel Award at the NCI-CCR Fellow's retreat, 2005

Preceptor for a Postdoctoral Fellow (T. Stewart), whose studies resulted in recognition with a competitive Travel Award at the NCI-CCR Fellow's retreat, 2005

Preceptor for a Research Fellow (K. Liu), whose studies resulted in recognition with an AACR-sponsored Scholar-in-Training Award to a special conference entitled "Oncogenomics: Dissecting Cancer through Genomic Research", 2005

Preceptor for a Postdoctoral Fellow (T. Stewart), who was selected for oral presentation at the Experimental Biology Conference, April 2004 in the Block Symposium on Antitumor Effector Cells, Mechanisms of Tumor Rejection and Modulation of Tumor Immunity

Preceptor for a Postdoctoral Fellow (S. Caldwell), who was selected for oral presentation at the NIH Immunology Retreat, 2003

Preceptor for a Research Fellow (K. Liu), who was selected for oral presentation at the NCI-CCR Fellow's Retreat, 2003

Preceptor for a Postdoctoral Fellow (S. Caldwell), who was selected for oral presentation at the NCI-CCR Fellow's Retreat, 2003

Preceptor for a Postdoctoral Fellow (M. Ryan), who was selected for oral presentation at the NCI-DBS Fellow's Retreat, 2000

Preceptor for a Research Fellow (E. Bergmann-Leitner), whose studies resulted in recognition with a NIH Fellows Award for Research Excellence (FARE), 1999

Preceptor for a Postdoctoral Fellow (J. A. Bristol), whose studies resulted in recognition with a NIH Fellows Award for Research Excellence (FARE), 1997

Teaching Activities (at RPCI and UB-SUNY)

Molecular Immunology (MIR511)
Trends in Tumor Immunology (MIR509)
Interferons and Cytokines (BIR530)
Advanced Topics in Immunology (MIR508)
Co-Organizer/Co-Chair of Student Journal Club (MIR522 throughout academic year)
Organizer of Departmental Data Club
Basics in Grantsmanship (MIR510)
Fundamentals of Immunology (MIC 512/412)
Oncology for Scientists (RPN530)

Invitations to Speak

Invited Speaker at the 2nd International Conference on Vaccines and Vaccination, Chicago, IL, August 20, 2012. Title of Seminar: "Mechanisms of Tumor-Induced Myeloid-Derived Suppressor Cell Development"

Invited Speaker to the Molecular and Developmental Genetics Seminar Series, Department of Molecular and Cellular Biology, Roswell Park Cancer Institute, Buffalo, NY, December 4, 2008. Title of Seminar: "Role of Interferon Regulatory Factor-8 (IRF-8) in Host-Tumor Immunosurveillance"

Invited Speaker at the 11th Annual Upstate New York Immunology Conference, Albany Medical College, Bolton Landing, NY, October 19-22, 2008. Title of Seminar: "IRF-8 Regulates Tumor Cell Responses to Apoptosis and Host Immunosurveillance"

Invited Speaker at the 8th Annual Buffalo Conference on Immunology, Buffalo, NY, September 8 – 9, 2008. Title of Seminar: "Role of IRF-8 in Tumor-Cell Response to Immunosurveillance Mechanisms"

Invited Speaker to the Tumor Immunology Seminar Series, Department of Immunology, Roswell Park Cancer Institute, Buffalo, NY, May 27, 2008. Title of Seminar: "Role of Interferon Regulatory Factor-8 in Myeloid-Derived Suppressor Cell Development and Function"

Invited Speaker to the Research Seminar Series, Dept. of Pediatrics, University of Texas M.D. Anderson Cancer Center, August 2007. Title of Seminar: "Fas Resistance Contributes to Tumor Escape and Progression"

Invited Speaker to The GW Cancer Center and the McCormick Center, in conjunction with the Department of Microbiology and Immunology, George Washington University Medical Center, November 2006. Title of Presentation: "Resistance to Fas-Mediated Lysis as a Mechanism of Immune Selection and Tumor Progression"

Invited Speaker to the Tumor Vaccine and Cell Therapy Working Group, AACR, Washington, D.C., April 2006. Title of Presentation: "Adoptive Immunotherapy Mediates Tumor Regression and Tumor Escape"

Invited Speaker to the Translational Immunology Seminar Series, Section of General Surgery, the University of Chicago Medical Center, November 2005. Title of Seminar: "Fas-Mediated Cytotoxicity as a Mechanism of Immunoselection and Tumor Escape"

Major Invited Speaker at an International Conference, entitled "Cancer Vaccines/Adjuvants & Delivery Systems for the Next Decade", Lisbon, Portugal, September 2005. Title of Presentation: "CTL-Based Immunotherapy Mediates Tumor Regression and Tumor Escape"

Invited Speaker, Hematology Branch, NHLBI, NIH, July 2005. Title of Seminar: "Combinatorial Approaches involving Immunotherapy and Radiation Enhance Tumor Regression"

Invited Speaker, Immunology Faculty, NCI, February 2005. Title of Seminar: "Positive and Negative Consequences of Fas/Fas Ligand Interactions in the Antitumor Response"
Major Invited Speaker at the University of Colorado for a Symposium on Cancer Biology,

November 2004. Title of Presentation: “Fas/Fas Ligand Interactions in the Regulation of Tumor Progression”

Major Invited Speaker at the University of Vermont for a Symposium on The Course of Cancer, October 2004. Title of Presentation: “Interactions Between the Cellular Immune System and Cancer”

Invited Speaker at the Gerber Adult Seminar series on Science & Technology, Rockville, MD, November 2004. Title of Seminar: “The Development of Vaccines for Cancer

Invited Speaker at the Breast Cancer Faculty, CCR, NCI, October 2004. Title of Seminar: “Immunologic Characteristics of a Transgenic Mouse Model of Primary and Metastatic Mammary Carcinoma”

Invited Speaker at the “8th World Congress on Advances in Oncology and 6th International Symposium on Molecular Medicine” in Crete, Greece, October 2003. Title of Presentation: “Regulation of the Fas Pathway in Tumor Immunotherapy”

Invited Speaker at the Regina Elena Cancer Institute in Rome, Italy, October 2003. Title of Seminar: “Combinatorial Vaccine Strategies Employing Recombinant Vectors”

Invited as a Keynote Speaker at an international conference on “Biotherapy of Cancer: From Disease to Targeted Treatment” in Munich, Germany, September 2003. Title of Presentation: “Combinatorial Vaccine Strategies Employing Recombinant Vectors”

Guest Lecturer in the Tumor Biology Program at the Georgetown University School of Medicine, April 2003. Title of Lecture: "Understanding the Host/Tumor Interaction for Cancer Vaccine Development”

Invited Speaker for the NCI-Frederick Seminar Series, November 2002. Title of Seminar: “Fas-Based Interactions in Tumor Regression and Progression”

Guest Lecturer in the Tumor Biology Program at the Georgetown University School of Medicine, April 2002. Title of Lecture: "Understanding the Host/Tumor Interaction for Cancer Vaccine Development”

Invited Speaker at the 11th International Congress of Immunology, Stockholm, July 2001. Title of Presentation: “Role of Fas in Human Antigen-Specific Cytotoxic T Lymphocyte-Colon Carcinoma Cell Interactions”

Invited Speaker in the Lombardi Cancer Center's Tumor Biology Seminar Series at the Georgetown University School of Medicine, October 2000. Title of Seminar: Regulation of the Fas/FasL Pathway in Human CD8+ Cytotoxic T Lymphocyte-Colon Carcinoma Cell Interactions”

Invited Speaker in the Cancer Center Distinguished Lecture Series at the Medical College of Wisconsin, July 2000. Title of Seminar: “ras Oncogene Products as Targets for Tumor Immunotherapy”

Guest Lecturer, “Immunotherapy of Cancer”, George Washington University School of Medicine, Molecular and Cellular Oncology Program, November 1999

Invited Speaker to the Department of Microbiology and Immunology, Indiana University School of Medicine, May 1998. Title of Seminar: “ras Oncogenes as Targets for Cancer Immunotherapy”

Invited Speaker at the Conference for Immunology and Immunotherapy of Metastasis, Title of Presentation: “Recombinant Vaccines to Point-Mutated ras and CEA: Analyses of Patient T-Cell Responses”, Lake Tahoe, CA, May1996. Also, Chairperson of one of the scientific session at this same meeting, entitled: “Immunotherapy of Metastasis”

Invited Speaker at the First International Conference on Gene Therapy & Vaccines for Cancer, Washington, D.C., 1994. Title of Presentation: “Peptides Reflecting Mutated ras p21 Epitopes Induce Host Cellular Immune Responses”

Invited Speaker to the Department of Pathology, “Immunology Seminar Program” at The Ohio State University College of Medicine, January 1994. Title of Seminar: “Induction and Characterization of Host Cellular Immune Responses to Mutated ras p21 Epitopes”

Ongoing Research Support

Source: DOD W81XWH-11-1-0394
Title: Granulopoietic Growth Factor Secretion in Ovarian Carcinoma as a Mechanism for the Emergence of Immune Suppressive Myeloid Subsets
Role: Principal Investigator
Duration of funding: 5/1/11 - 4/30/13
Goals of Study: This research tests *the novel hypothesis that human ovarian tumor cells produce the granulopoietic growth factor, G-CSF that participates in the generation of granulocytic MDSC.* This hypothesis will be tested by pursuing three aims: *1)* to quantify G-CSF levels in human ovarian cancer cell lines, and then evaluate the ability of selected G-CSF-producing cell lines to generate MDSC using human-mouse xenograft models; *2)* to examine the causal link between tumor-derived G-CSF production and granulocytic MDSC development using loss-of-function approaches; and *3)* to measure circulating G-CSF and MDSC levels in ovarian carcinoma patients and then correlate potential changes in both parameters with clinical outcome measurements.
Overlap: None

Source: NCI/NIH R01CA140622
Title: IRF-8 as a Negative Regulator of $CD11b^{+}Gr\text{-}1^{+}$ Myeloid Cell Production and Function
Role: Principal Investigator
Duration of Funding: 9/1/11 – 7/31/16
Goals of Study: This research tests *the novel hypothesis that such dysfunctional myeloid populations accumulate or become pro-tumorigenic because neoplastic cells cause a profound alteration in interferon regulatory factor-8 (IRF-8) that is normally essential for controlling fundamental properties of the myeloid cell family*. This hypothesis will be tested by pursuing three aims: *1)* to demonstrate a causal link between IRF-8 expression and tumor-induced $CD11b^{+}Gr\text{-}1^{+}$ myeloid cell development/function; *2)* to identify the molecular mechanisms of IRF-8 regulation; and *3)* to investigate the therapeutic value of IRF-8 enhancement against tumor growth.
Overlap: None

Completed Research Support

Source: Alliance Developmental Award from the Roswell Park Alliance Foundation
Title: Development of Myeloid-Derived Suppressor Cells in Mammary Carcinoma through Interferon Regulatory Factor-8-Dependent Mechanisms
Role: Principal Investigator
Duration of funding: 11/1/10 – 10/31/11
Goals of Study: RPCI Alliance Foundation has awarded seed funding to explore the relationship between MDSC accumulation/IRF-8 expression and disease status or outcome in patients with breast cancer.

Source: NO8G-370 - NYSTEM (New York State Stem Cell Science)
Title: Role of Cancer Stem Cells in Resistance to Targeted Therapy and Tumor Recurrence
Role: Co-Investigator
Duration of funding: 1/1/09 – 6/30/11
Goals of Study: NYSTEM has provided seed funding for an exploratory study investigating the sensitivity of pancreatic cancer stem cells to novel death-inducing agents. This grant proposal is designed to generate data on the apoptotic sensitivity of pancreatic cancer stem cells to Apo2L/TRAIL from a large cohort of pancreatic patient tumors both *in vitro* and *in vivo* in xenograft models.

Source: Alliance Developmental Award from the Roswell Park Alliance Foundation
Title: Regulation of Interferon Regulatory Factor-8 in Neoplastic Cells to Augment Responses to Apoptosis and Immunotherapy
Role: Principal Investigator
Duration of funding: 1/1/09 – 12/31/09 *Goals of Study*: The goal of this study was to explore the hypothesis that histone deacetylase inhibitors (HDACi), a novel class of epigenetic modifiers, enhance Fas-mediated apoptosis through IRF-8-dependent mechanisms. Our findings revealed that HDACi upregulated IRF-8 expression in tumor cells, which resulted in enhanced Fas-mediated apoptosis. Tumor cells rendered IRF-8-deficient were significantly less susceptible to Fas-mediated apoptosis, demonstrating that IRF-8 was required for HDACi-mediated antitumor activity. Therefore, from a clinical perspective, IRF-8 status may help to explain the relative efficacy of certain types of epigenetic therapies.

Publications

1. Bray, R., **Abrams, S.**, and Brahmi, Z. Studies on the mechanism of human natural killer cell-mediated cytolysis. I. Modulation by dexamethasone and arachidonic acid. Cell. Immunol. 78: 100-113, 1983.

2. **Abrams, S. I.**, Bray, R. A., and Brahmi, Z. Mechanism of action of phorbol myristate acetate on human natural killer cell activity. Cell. Immunol. 80: 230-40, 1983.

3. **Abrams, S. I.**, and Brahmi, Z. Compared mechanisms of tumor cytolysis by human natural killer cells and activated polymorphonuclear leukocytes. J. Immunol. 132: 3192-3196, 1984.

4. Brahmi, Z., Bray, R. A., and **Abrams, S. I.** Evidence for an early calcium-independent event in the activation of the human natural killer cell cytolytic mechanism. J. Immunol. 135: 4108-4113, 1985.

5. **Abrams, S. I.**, and Brahmi, Z. The functional loss of human natural killer cell activity induced by K562 is reversible via an interleukin-2-dependent mechanism. Cell. Immunol. 101: 558-570, 1986.

6. **Abrams, S. I.**, and Brahmi, Z. Target cell directed NK inactivation. Concomitant loss of NK and antibody-dependent cellular cytotoxicity activities. J. Immunol. 140: 2090-2095, 1988.

7. **Abrams, S. I.**, and Brahmi, Z. Mechanism of K562-induced human natural killer cell inactivation using highly enriched effector cells isolated via a new single-step sheep erythrocyte-rosette assay. Ann. Inst. Pasteur/Immunol. 139: 361-381, 1988.

8. **Abrams, S. I.**, McCulley, D. E., Meleedy-Rey, P., and Russell, J. H. Cytotoxic T lymphocyte-induced loss of target cell adhesion and lysis involve common and separate signaling pathways. J. Immunol. 142: 1789-1796, 1989.

9. **Abrams, S. I.**, and Russell, J. H. $CD4^+$ T lymphocyte-induced target cell detachment. A model for T cell-mediated lytic and nonlytic inflammatory processes. J. Immunol. 146: 405-413, 1991.

10. Russell., J. H., Rush, B. J., **Abrams, S. I.**, and Wang, R. Sensitivity of T-cells to anti-CD3-stimulated suicide is independent of functional phenotype. Eur. J. Immunol. 22: 1655-1658, 1992.

11. Kantor, J., Irvine, K., **Abrams, S.**, Kaufman, H., DiPietro, J., and Schlom, J. Antitumor activity and immune responses induced by a recombinant carcino-embryonic antigen-vaccinia virus vaccine. J. Natl. Cancer Inst. 84: 1084-1091, 1992.

12. Kantor, J., Irvine, K., **Abrams, S.**, Snoy, P., Olson, R., Greiner, J., Kaufman, H., Eggensperger, D., and Schlom, J. Immunogenicity and safety of a recombinant vaccinia virus vaccine expressing the carcinoembryonic antigen gene in a nonhuman primate. Cancer Res. 52: 6917-6925, 1992.

13. **Abrams, S. I.**, Wang, R., Munger, W. E., and Russell, J. H. Detachment and lysis of adherent target cells by $CD4^+$ T cell clones involve multiple effector mechanisms. Cell. Immunol. 147: 188-202, 1993.

14. Wang, R., **Abrams, S. I.**, Loh, D., Hsieh, C-S., Murphy, K. M., and Russell, J. H. Separation of $CD4^+$ functional responses by peptide dose in T_h1 and T_h2 subsets expressing the same transgenic antigen receptor. Cell. Immunol. 148: 357-370, 1993.

15. Kantor, J., **Abrams, S.**, Irvine, K., Snoy, P., Kaufman, H., and Schlom, J. Specific immunotherapy using a recombinant vaccinia virus expressing human carcino-embryonic antigen. Ann. N.Y. Acad. Sci. 690: 370-373, 1993.

16. Conry, R. M., LoBuglio, A. F., Kantor, J., Schlom, J., Loechel, F., Moore, S. E., Sumerel, L. A., Barlow, D .L., **Abrams, S**., and Curiel, D. T. Immune response to a carcino-embryonic antigen polynucleotide vaccine. Cancer Res. 54: 1164-1168, 1994.

17. Bei, R., Kantor, J., Kashmiri, S. V. S., **Abrams, S**., and Schlom, J. Enhanced immune responses and anti-tumor activity by baculovirus recombinant CEA in mice primed with the recombinant vaccinia CEA. J. Immunother. 16: 275-282, 1994.

18. Hodge, J. W., **Abrams, S.**, Schlom, J., and Kantor, J. A. Induction of antitumor immunity by recombinant vaccinia viruses expressing B7-1 or B7-2 costimulatory molecules. Cancer Res. 54: 5552-5555, 1994.

19. Hodge, J. W., McLaughlin, J. P., **Abrams S. I.**, Shupert, W. L., Schlom, J., and Kantor, J. A. Admixture of a recombinant vaccinia virus containing the gene for the costimulatory molecule B7 and a recombinant vaccinia virus containing a tumor associated antigen gene results in enhanced specific T-cell responses and antitumor immunity. Human Gene Ther. 8: 851-860, 1997.

25. Prasad, G. L., Lee, H-S., Iwahashi, M., Milenic, D., **Abrams, S.**, Schlom, J., and Kashmiri, S. *In vivo* gene inoculation of a recombinant single-chain antitumor antibody induces anti-immunoglobulin response. Cancer Gene Ther. 4: 253-259, 1997.

26. McLaughlin, J. P., **Abrams, S.**, Kantor, J., Dobrzanski, M. J., Greenbaum, J., Schlom, J., and Greiner, J. W. Immunization with a syngeneic tumor infected with recombinant vaccinia virus expressing granulocyte-macrophage colony-stimulating factor (GM-CSF) induces tumor regression and long-lasting systemic immunity. J. Immunother. 20: 449-459, 1997.

27. **Abrams, S. I.**, Khleif, S. N., Bergmann-Leitner, E. S., Kantor, J. A., Chung, Y, Hamilton, J. M., and Schlom, J. Generation of stable $CD4^+$ and $CD8^+$ T cell lines from patients immunized with *ras* oncogene-derived peptides reflecting codon 12 mutations. Cell. Immunol. 182: 137-151, 1997.

28. Bristol, J. A., Schlom, J., and **Abrams, S. I.** Development of a murine mutant *ras* $CD8^+$ CTL peptide epitope variant that possesses enhanced MHC class I binding and immunogenic properties. J. Immunol. 160: 2433-2441, 1998.

29. Bergmann-Leitner, E. S., Kantor, J. A., Shupert, W. L., Schlom, J., and **Abrams, S. I.** Identification of a human $CD8^+$ T lymphocyte neo-epitope created by a *ras* codon 12 mutation restricted by the HLA-A2 allele. Cell. Immunol. 187: 103-116, 1998.

30. Bristol, J. A., Schlom, J., and **Abrams, S. I.** Persistence, immune specificity, and functional ability of murine mutant *ras* epitope-specific $CD4^+$ and $CD8^+$ T lymphocytes following *in vivo* adoptive transfer. Cell. Immunol. 194: 78-89, 1999.

31. Khleif, S. N., **Abrams, S. I.**, Hamilton, J. M., Bergmann-Leitner, E., Chen, A., Bastian, A., Bernstein, S., Chung, Y., Allegra, C. J., and Schlom, J. A phase I vaccine trial with peptides reflecting *ras* oncogene mutations of solid tumors. J. Immunother. 22: 155-165, 1999.

32. Bergmann-Leitner, E. S., and **Abrams, S. I.** Differential role of Fas/Fas ligand interactions in cytolysis of primary and metastatic colon carcinoma cell lines by human antigen-specific $CD8^+$ CTL. J. Immunol. 164: 4941-4954, 2000.

33. Bergmann-Leitner, E. S., and **Abrams, S. I.** Influence of interferon-□ on modulation of Fas expression by human colon carcinoma cells and their subsequent sensitivity to antigen-specific $CD8^+$ cytotoxic T lymphocyte attack. Cancer Immunol. Immunother. 49: 193-207, 2000.

34. Bristol, J. A., Orsini, C., Lindinger, P., Thalhamer, J., and **Abrams, S. I.** Identification of a *ras* oncogene peptide that contains both $CD4^+$ and $CD8^+$ T cell epitopes in a nested configuration and elicits both T cell subset responses by peptide or DNA immunization. Cell. Immunol. 205: 73-83, 2000.

35. Ryan, M. H., and **Abrams, S. I.** Characterization of $CD8^+$ cytotoxic T lymphocyte-tumor cell interactions reflecting recognition of an endogenously expressed murine wild-type p53 determinant. Cancer Immunol. Immunother. 49: 603-612, 2001.

36. Alper, O., Bergmann-Leitner, E. S., **Abrams, S.**, and Cho-Chung, Y. S. Apoptosis, growth arrest and suppression of invasiveness by CRE-decoy oligonucleotide in ovarian cancer cells: Protein kinase A downregulation and cytoplasmic export of CRE-binding proteins. Mol. Cell. Biochem. 218: 55-63, 2001.

37. Bergmann-Leitner, E. S., and **Abrams, S. I.** Positive and negative consequences of soluble Fas ligand produced by an antigen-specific $CD4^+$ T cell response in human carcinoma immune interactions. Cell. Immunol. 209: 49-62, 2001.

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41. Chakraborty, M., **Abrams, S. I.**, Camphausen, K., Liu, K., Scott, T., Coleman, C. N., and Hodge, J. W. Irradiation of tumor cells upregulates Fas, enhances CTL lytic activity and CTL adoptive immunotherapy. J. Immunol. 170: 6338-6347, 2003.

42. Liu, K., and **Abrams, S. I.** Alterations in Fas expression are characteristic of, but not solely responsible for, enhanced metastatic competence. J. Immunol. 170: 5973-5980, 2003.

43. Caldwell, S. A., Ryan, M. H., McDuffie, E., and **Abrams, S. I.** The Fas/Fas ligand pathway is important for optimal tumor regression in a mouse model of CTL adoptive immunotherapy of experimental CMS4 lung metastases. J. Immunol. 171: 2402-2412, 2003.

44. Lindinger, P., Mostböck, S., Hammerl, P., Hartl, A., Thalhamer, J., and **Abrams, S. I.** Induction of murine *ras* oncogene peptide-specific T cell responses by immunization with plasmid DNA-based minigene vectors. Vaccine 21: 4285-4296, 2003.

45. Liu, K., McDuffie, E., and **Abrams, S. I.** Exposure of human primary colon carcinoma cells to anti-Fas interactions influences the emergence of pre-existing Fas-resistant metastatic subpopulations. J. Immunol. 171: 4164-4174, 2003.

46. Chakraborty, M., **Abrams, S. I.**, Camphausen, K., Coleman, C. N., Schlom, J., and Hodge, J. W. External beam radiation of tumors alters phenotype of tumor cells to render them susceptible to vaccine-mediated T-cell killing. Cancer Res. 64: 4328-4337, 2004.

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49. Nesterova, M. V., Johnson, N., Stewart, T., **Abrams, S**., and Cho-Chung, Y. S. CpG-immunomer DNA enhances antisense protein kinase A RI . Inhibition of multidrug-resistant colon carcinoma growth in nude mice: molecular basis for combinatorial therapy. Clin. Cancer Res. 11: 5950- 5955, 2005.

50. Liu, K., Caldwell, S. A., Greeneltch, K. M., Yang, D., and **Abrams, S. I.** CTL adoptive immunotherapy concurrently mediates tumor regression and tumor escape. J. Immunol. 176: 3374- 3382, 2006.

51. Gelbard, A., Garnett, C. T., **Abrams, S. I.**, Patel, V., Gutkind, J. S., Palena, C., Tsang, K-Y., Schlom, J., and Hodge, J. W. Combination chemotherapy and radiation of human

squamous cell carcinoma of the head and neck augments CTL-mediated lysis. Clin. Cancer Res. 12: 1897-1905, 2006.

52. Lundqvist, A., **Abrams, S. I.**, Schrump, D. S., Alvarez, G., Suffredini, D., Berg, M., and Childs, R. Bortezomib and depsipeptide sensitize tumors to tumor necrosis factor-related apoptosis-inducing ligand: a novel method to potentiate natural killer cell tumor cytotoxicity. Cancer Res. 66: 7317-7325, 2006.

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54. Yang, D., Thangaraju, M., Greeneltch, K., Browning, D., Schoenlein, P., Tamura, T., Ozato, K., Ganapathy, V., **Abrams, S. I**., Liu, K. Repression of IFN regulatory factor 8 by DNA methylation is a molecular determinant of an apoptotic resistance and metastatic phenotype in metastatic tumor cells. Cancer Res. 67: 3301-3309, 2007.

55. Stewart, T. J., and **Abrams, S. I**. Altered immune function during long-term host-tumor interactions can be modulated to retard autochthonous neoplastic growth. J. Immunol. 179: 2851-2859, 2007.

56. Yang, D., Din, N., Browning, D. D., **Abrams, S. I**., and Liu, K. Targeting lymphotoxin β receptor with tumor-specific T lymphocytes for tumor regression. Clin. Cancer Res. 13: 5202- 5210, 2007.

57. Greeneltch, K. M., Schneider, M. Steinberg, S. M., Liewehr, D. J., Stewart, T. J., Liu, K., and **Abrams, S. I**. Host immunosurveillance controls tumor growth via IRF-8-dependent mechanisms. Cancer Res. 67: 10406-10416, 2007.

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60. Stewart TJ, **Abrams SI**. How tumours escape mass destruction. Oncogene. 27: 5894-5903, 2008.

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Stewart TJ, Liewehr DJ, Steinberg SM, Greeneltch KM, **Abrams SI**. Modulating the expression of interferon regulatory factor-8 alters the pro-tumorigenic behavior of $CD11b^{+}Gr\text{-}1^{+}$ myeloid cells. J. Immunol. 183: 117-128, 2009. PMC2744444

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64. Godoy H, Kesterson J, Eppolito C, Vethanayagam RR, Grimm M, **Abrams S**, Lele S, Segal B, Odunsi A. NADPH Oxidase Modulates the Development of Myeloid-Derived Suppressor Cells (MDSCs) in Ovarian Cancer Tumor-Bearing Hosts. Reprod. Sci. 18: 290A, 2011.

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Book Chapters

1. Russell, J. H., and **Abrams, S. I.** Target cell events initiated by T cell attack. In: Cytotoxic Cells: Recognition, Effector Function, Generation, and Methods,Sitkovsky, M. and Henkart, P. (Eds.), Birkhauser, Boston, 202-212, 1993.

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